

A comparative study on two extraction procedures in speciation of iron in municipal solid waste

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ABSTRACT

Two extraction reagents, hydrochloric acid (HCl) and acid ammonium oxalate solution (Tamm's reagent), were used to evaluate the redox state of iron in municipal solid waste (MSW) with different deposit ages. Orthogonal experiments were conducted to optimize the extraction conditions for extractable iron speciation (ferric and ferrous) in MSW. The optimal extraction conditions for HCl were determined as follows: the liquid-to-solid ratio was set at 100, and then the samples were extracted at the shaking speed of 200 rpm at 35 °C for 60 min by 1.00 M HCl. For Tamm's reagent, the optimal extraction conditions were extracted at the shaking speed of 175 rpm at 30 °C for 12 h with the same liquid-to-solid ratio. However, Tamm's reagent extraction is much more laborious and time-consuming. Thus the HCl extraction might be a better choice for the evaluation of the redox state of iron in MSW. The results also showed that the yield of extractable iron increased with deposited age. About 60–83% of extractable iron was presented as ferrous in the MSW deposited for 1–8 years. This study supplied a tool for investigating the role of iron on the fate of pollutants in the landfill.

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1. Introduction

Landfill as municipal solid waste (MSW) disposal is a widely accepted technology, especially in developing countries. Iron is the fourth-most abundant element in the earth's crust and widely used in the production and life. Therefore, it is the most prevalent heavy metal in the MSW and leachate [1]. For instance, Flyhammar [2] showed that the total amount of iron in the fresh MSW and degraded waste were up to 12,400 and 10,300 mg kg⁻¹, respectively. Kjeldsen et al. [3] summarized the ranges of general leachate parameters from various reports, and they found iron concentration in the leachate varied from 3 to 5500 mg L⁻¹. Oman and Junestedt [4] showed that iron concentration in the leachate varied more than 250 times between 0.16 and 42.7 mg L⁻¹ while the iron in the leachate sediment varied between 13,500 and 285,000 mg kg⁻¹. However, most of environmental scientists just paid attention on the visual pollution resulted from leachate iron, and very few have

shown their concern on the role of the iron on the migration and transformation of inorganic and organic pollutants in the landfill.

In most soils and sediments, iron is dominant redox-active metal [5]. Geomicrobiological cycling of iron has a strong influence on the carbon, sulfur and nitrogen cycles [1], and the valence of iron in the environment is a good indicator of redox condition and microbial activity. Under various anoxic environmental conditions, such as soils and sediments, dissimilatory of ferric iron reduction is an important microbial process for organic matter degradation [6,7]. The produced ferrous iron can be reoxidized by biological as well as abiological process, but the latter one can only be available under extreme conditions and is believed to be of little significance [8]. Hence, the oxidation process of ferrous iron in anoxic condition could be mainly catalyzed by microorganisms, such as phototrophic and nitrate-reducing bacteria [9–11]. Nowadays, impacts of iron cycle under anoxic environment on the fate of both organic and inorganic pollutants have been investigated in the soil and sediment [12–14]. As was mentioned above, there were huge amounts of iron in the MSW and leachate. Therefore, microbial anaerobic iron cycling process in the landfill is also theoretical feasible and would also play an important role on the degradation of organic and inorganic pollutants.

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In order to evaluate the importance of iron on the degradation of organic matters and other pollutants in the landfill, some suitable methods are required to study the different form of iron like ferric and ferrous. Unfortunately, to our knowledge, no such research have been conducted with MSW, whereas numerous studies have been performed in soil [15,16], sediment [6,17–20] and activated sludge [21] using various extraction methods. Importantly, unlike soil, sediment and activated sludge, the constituent and basic physicochemical characteristics of MSW were significantly influenced by the deposited age [22]. Moreover, since MSW was an extremely heterogeneous substrate independent of its geometry, particle size or chemical composition [2], and high content of organic matter and the heterogeneity of MSW made it obviously different from soil, sediment and activated sludge. Therefore, the speciation of the redox state of iron to different aged refuse studies would be advantageous, and the development of such an extraction technique might ultimately enable us to get a better understanding on the role of iron during the degradation process of MSW.

The aim of the present study was to find a feasible extraction method for characterization and quantitative analysis of ferrous (Fe(II)), ferric (Fe(III)) and total extractable iron contents in MSW. Due to no reference samples for MSW was available till now, the methods optimization were carried by the fortified recovery. Moreover, in order to provide detail information on the speciation of redox state, MSW of different deposit ages were investigated by evaluating the effect of different extract procedures on the dissolution of ferrous and ferric irons.

2. Materials and methods

2.1. Reagents

All the reagents used in our experiment were of analytical reagent grade unless otherwise stated. All dissolutions and dilutions were performed with distilled water and all the plastic and glassware were cleaned up by soaking dilute HNO_3 (Zhongxing Chemical Reagent Co., Ltd, Zhejiang, China).

Hydrochloric acid (HCl) extractant, 1.0 M, was prepared from HCl (Hangzhou Chemical Reagent Co., Ltd, Zhejiang, China). Other concentrations of HCl, such as 0.25, 0.5 and 0.75 M, were obtained by diluted 1 M HCl solution.

The acid ammonium oxalate solution (Tamm's reagent), namely 0.175 M $(\text{NH}_4)_2\text{C}_2\text{O}_4 \cdot \text{H}_2\text{O}$ -0.100 M $\text{H}_2\text{C}_2\text{O}_4 \cdot 2\text{H}_2\text{O}$, was prepared from ammonium oxalate (Meixing Chemical Reagent Co., Ltd, Shanghai, China) and oxalic acid (Shishihewei Chemical Reagent Co., Ltd, Shanghai, China). In order to remove oxygen in the solution, Tamm's reagent was bubbled with N_2 which passed through a heated column of reduced copper filings and closed with stopper. Ferrous stock standard solution, 100 mg L^{-1} , was prepared from $\text{Fe}(\text{NH}_4)_2(\text{SO}_4)_2 \cdot 6 \text{H}_2\text{O}$ (Huzhou Chemical Reagent Co., Ltd, Zhejiang, China). *o*-Phenanthroline, 0.5%, prepared from $\text{C}_{12}\text{H}_8\text{N}_2 \cdot \text{H}_2\text{O}$ (Huaqiao Chemical Reagent Co., Ltd, Zhejiang, China).

Sodium acetate buffer of pH 5.0 was prepared from sodium acetate (Wenzhou Chemical Reagent Co., Ltd, Zhejiang, China) and acetic acid (Hangzhou Chemical Reagent Co., Ltd, Zhejiang, China).

Standards stock solution for the flame atomic absorption spectrometer assay (1000 mg L^{-1}) was purchased from National Analysis Center for Iron and Steel (NACIS) (Beijing, China).

Amorphous Fe(III) oxyhydroxide was prepared by neutralizing 0.4 M FeCl_3 (Sinopharm Chemical Reagent Co., Ltd, Shanghai, China) to pH 7 with 1.0 M NaOH (Zhongxing Chemical Reagent Co., Ltd, Zhejiang, China) and washing the suspension with deionized water to remove the NaCl [5]. The total amount of iron in the amorphous Fe(III) oxide was determined by dissolving the Fe(III) oxide in concentrated HCl, diluting the solutions with deionized water, and assaying for iron by flame AAS (Thermo iCE-3500).

2.2. Sampling and pretreatment

MSW samples were collected from Tianziling MSW landfill site, which is located in a valley north of Hangzhou City ($30^\circ 23' \text{N}$, $120^\circ 12' \text{E}$), Zhejiang province, China. Its bottom liners located at 54 m above sea level and the top of the landfill will reach 165 m above sea level. The matured refuse was located at bottom deposited layers and the fresh one at top deposit layers. This facility initiated its operation in April 1991. Up to the end of 1996, the first stage of the project had disposed of around 8 million tones of refuse and was closed. The second stage of the project was constructed in December 2003 and started operation in 2006, occupying a total area of about 113 ha, and the waste loading is about $1940\text{--}4000 \text{ t day}^{-1}$.

Zero year aged refuse (fresh refuse), sample S_0 were collected from a working flat at the top layer of landfill site. The other four MSW samples were collected from four different deposit layers in a sampling well drilled, the sampling procedure as follows. First of all, two wells of depth 15 m (well 1) and 20 m (well 2) were drilled at the landfill site. After then, the samples were collected at depths of 8 m, 15 m in well 1, 5 m and 20 m in the well 2. According to the landfill records of waste placement, the four sampling layers were with the deposit age of 1(S_1), 2(S_2), 4(S_4) and 8(S_8) year respectively. At least 2–5 kg sub-samples for each were taken. Samples were immediately placed in an airtight plastic bag during the collection and filled with nitrogen gas N_2 , and then sealed. All samples were transported to the laboratory where they were frozen until analysis. Larger inert objects (including stones, pieces of brick, concrete and cinders) were removed, and then each sample was quartered to $\sim 500 \text{ g}$ sub-samples under the stream of N_2 , which passed through a heated column of reduced copper filings to remove any traces of oxygen. A part of them ($\sim 200 \text{ g}$) was used to determine the pH, biodegradable matter (BDM), moisture and ash content. The rest of the sample ($\sim 300 \text{ g}$) was grinded and homogenized under the stream of N_2 as evenly as possible, and then kept in our lab under anaerobic conditions.

$S_{0.5}$, with the deposit age of 0.5 years, was also collected from the same landfill site at depths of 2 m in the well 2. After removed larger inert objects, $S_{0.5}$ was air dried at 30°C for 15 days. Subsequently, all the air-dried samples were grinded and sieved through a 1 mm nylon mesh and then homogenized with quadrant division method as depicted by Long et al. [23]. As was mentioned before, the MSW was an extremely heterogeneous substrate. Thus, for the sake of eliminating the impact of heterogeneous on the optimization experiment, the MSW sample of $S_{0.5}$ was chosen for orthogonal test. After homogenization, the moisture content of $S_{0.5}$ was adjusted to 30% by distilled water and then kept under anaerobic conditions for 30 days before the orthogonal test started.

2.3. Experimental design for optimization of extraction parameters

2.3.1. Optimization of iron extraction

The optimizations of hydrochloric acid (HCl) and Tamm's reagent extraction were conducted by orthogonal test. Different concentrations HCl of have been used to dissolve acid-soluble or cold-extractable metals from soils [15], sediments [24,25] and activated sludge [21]. However, according to the extraction results reported by the authors, who had used HCl to extract iron from sediments, soils and activated sludge, the extraction process was affected by various factors, such as extraction time, extraction temperature, the concentration of HCl and etc. [15,20,21,24]. Therefore, according to the factor and level commonly used in the extraction of iron from soil, sediment and activated sludge by using HCl, five independent variables with four variation levels were considered in this study, namely A (liquid-to-solid ratio: 25, 50, 75 and

Table 1
Factors and levels for orthogonal test to the extractant of HCl.

Design variable	Levels			
	1	2	3	4
(A) Solution-to-sample ratio (n)	25	50	75	100
(B) HCl concentrations (M)	0.25	0.50	0.75	1.00
(C) Extraction time (min)	15	30	45	60
(D) Rotational speed (rpm)	125	150	175	200
(E) Extraction temperature (°C)	25	30	35	40

100), B (HCl concentration: 0.25, 0.50, 0.75, and 1.00 M), C (extraction time: 15, 30, 45 and 60 min), D (shaking speed: 125, 150, 175 and 200 rpm), and E (extraction temperature: 25, 30, 35 and 40 °C) (Table 1). The table of orthogonal array is formed by using the orthogonal array L₁₆ (4⁵). Table 2 showed the composition of the table of orthogonal array, and the extract result of the ferrous (Fe(II)), ferric (Fe(III)) and total extractable iron.

The acid ammonium oxalate solution (0.175 M (NH₄)₂C₂O₄–0.100 M H₂C₂O₄), commonly known as Tamm's reagent, has been extensively used in studies of soil genesis and exploration geochemistry as a selective extractant to dissolve amorphous iron oxides in darkness. Based on the extraction results reported by the authors, who had used Tamm's reagent to extract iron from sediments, soils and activated sludge, the extraction process was also affected by various factors, such as extraction time, extraction temperature and liquid-to-solid ratio and shaking speed [15,18,20,21,24]. Thus, four independent variables with three variation levels were selected based on the results which were commonly used in the extraction of iron from soil, sediment and activated sludge, namely A (liquid-to-solid ratio: 25, 50 and

Table 2
Analysis of L₁₆ (4⁵) test results with the extractant of HCl.

No.	A (n)	B (M)	C (min)	D (rpm)	E (°C)	Fe(II) (mg kg ⁻¹)	Fe(III) (mg kg ⁻¹)	Total (mg kg ⁻¹)
1	1	1	1	1	1	519	1280	1800
2	1	2	2	2	2	1280	1640	2920
3	1	3	3	3	3	2040	2220	4260
4	1	4	4	4	4	2090	3120	5210
5	2	1	2	3	4	1500	1500	3000
6	2	2	1	4	3	1970	3010	4990
7	2	3	4	1	2	2390	2860	5250
8	2	4	3	2	1	2220	3290	5510
9	3	1	3	4	2	1170	2740	3910
10	3	2	4	3	1	1990	2900	4900
11	3	3	1	2	4	2200	3070	5270
12	3	4	2	1	3	2530	3060	5580
13	4	1	4	2	3	1760	2530	4290
14	4	2	3	1	4	2130	2540	4670
15	4	3	2	4	1	2110	3840	5950
16	4	4	1	3	2	2080	2590	4680
<i>Fe (II)</i>								
K1	1480	1240	1690	1890	1710			
K2	2020	1840	1850	1860	1730			
K3	1970	2180	1890	1900	2080			
K4	2020	2230	2060	1840	1980			
R ^a	539	994	365	66	363			
<i>Fe (III)</i>								
K1	2070	2010	2490	2440	2830			
K2	2670	2520	2510	2630	2460			
K3	2940	3000	2700	2300	2700			
K4	2880	3010	2850	3180	2560			
R ^a	878	1000	361	872	369			
<i>Total</i>								
K1	3550	3250	4180	4330	4540			
K2	4690	4370	4360	4500	4190			
K3	4920	5180	4590	4210	4780			
K4	4900	5240	4910	5010	4540			
R ^a	1370	2000	727	806	590			

^a Refers to the result of extremedifference analysis.**Table 3**
Factors and levels for orthogonal test to the extractant of Tamm's reagent.

Design variable	Levels		
	1	2	3
(A) Solution-to-sample ratio (n)	25	50	100
(C) Extraction time (h)	6	12	24
(D) Rotational speed (rpm)	125	150	175
(E) Extraction temperature (°C)	25	30	35

100), B (extraction time: 6, 12 and 24 h), C (shaking speed: 125, 150 and 175 rpm), and E (extraction temperature: 25, 30 and 35 °C) (Table 3). The table of orthogonal array is formed by using the orthogonal array L₉ (3⁴). Table 4 showed the composition of the table of orthogonal array, and the extract result of the Fe(II), Fe(III) and total extractable iron.

2.3.2. Extraction of iron by HCl

The extraction can be performed under aerobic conditions and Fe(II) and Fe(III) will keep their original state rather than being oxidized or reduced during the extraction. In the present study, we introduced this extraction method to study the Fe(II) and Fe(III) extraction under anaerobic condition. Under a stream of N₂, which passed through a heated column of reduced copper filings to remove any traces of oxygen, fresh MSW sample (equal to 1 g dry sample) was transferred to a test tube containing hydrochloric acid, and then the tube was closed by stopper. Glass beads were added to aid stirring, and extraction took place on a shaker. The test tubes were centrifuged, the supernatant filtered (0.45 μm, Millipore filter) and the extractant analyzed for Fe(II) and total extractable iron as described below.

Table 4
Analysis of L_9 (3^4) test results with the extractant of Tamm's reagent.

No.	A (n)	C (h)	D (rpm)	E (°C)	Fe(II) (mg kg ⁻¹)	Fe(III) (mg kg ⁻¹)	Total (mg kg ⁻¹)
1	1	1	1	1	1600	2290	3890
2	1	2	2	2	1880	2670	4560
3	1	3	3	3	2150	2400	4550
4	2	1	2	3	1860	3320	5180
5	2	2	3	1	2290	2950	5230
6	2	3	1	2	1970	3400	5360
7	3	1	3	2	2040	3370	5410
8	3	2	1	3	2010	3360	5370
9	3	3	2	1	2330	3240	5570
<i>Fe (II)</i>							
K1	1880	1830	1860	2070			
K2	2040	2060	2030	1960			
K3	2130	2150	2160	2010			
R ^a	252	318	299	108			
<i>Fe (III)</i>							
K1	2460	2990	3010	2820			
K2	3220	2990	3080	3150			
K3	3320	3010	2910	3020			
R ^a	867	19	168	323			
<i>Total</i>							
K1	4330	4830	4870	4900			
K2	5260	5050	5100	5110			
K3	5450	5160	5070	5030			
R ^a	1120	336	229	215			

^a Refers to the result of extremedifference analysis.

2.3.3. Extraction of iron by Tamm's reagent

Fe(II) and total extractable iron were extracted by Tamm's reagent according to the procedures described by Phillips and Lovley [18], and Fe(II) was not oxidized and Fe(III) was not reduced when the extraction is performed under anaerobic conditions. Briefly, the solution of acid ammonium oxalate was bubbled with N₂, which was passed through a heated column of reduced copper filings to remove any traces of oxygen [26]. Under a stream of N₂, fresh MSW sample (equal to 1 g dry sample) was transferred to a serum bottle (150 ml), which was closed with chlorobutyl rubber stoppers and aluminum foil to exclude light. Tamm's reagent was injected into the serum bottle, and extraction took place in darkness on a shaker. After the extraction period, Fe(II) and total extractable iron were determined as described below.

2.4. Fe fortified recovery experiment

Based on the research results, the optimal extraction method was tested for the feasibility of characterizing and quantifying the Fe(II), Fe(III) and total extractable iron in different aged MSW. Since no reference samples for MSW were available, the only choice was fortified recovery to check the extraction techniques. Thus, for the sake of ensuring validity of the results, HCl and Tamm's reagent were tested for recovery of added Fe(II) and Fe(III). 0.05 M solutions of Fe(NH₄)₂(SO₄)₂·6H₂O and amorphous Fe(III) oxides were prepared for the fortified recovery of Fe(II) and Fe(III) either. According to the content of Fe(II) and Fe(III) in different aged refuse, 2.24–11.2 mg of Fe(II) or Fe(III) were added to fresh MSW samples (equal to 1 gram dry samples) of S₀, S₁, S₂, S₄ and S₈ under a stream of N₂, and the extraction proceeded as described above. The fortified recovery of Fe(II) and Fe(III) were calculated by equation (1) and (2). All the extractions and analyses were carried out in triplicate to ensure the validity of the results.

$$\text{Fe(II) recovery (\%)} = \frac{\text{Fe(II)}_y - \text{Fe(II)}_n}{\text{Fe(II)}_a} \times 100 \quad (1)$$

$$\text{Fe(III) recovery (\%)} = \frac{\text{Fe(III)}_y - \text{Fe(III)}_n}{\text{Fe(III)}_a} \times 100 \quad (2)$$

Where Fe(II)_y or Fe(III)_y represent the yields of Fe(II) or Fe(III) with added Fe(II) or synthetic amorphous Fe(III) oxide after extracted by HCl or Tamm's reagent, Fe(II)_n or Fe(III)_n represent the yields of Fe(II) or Fe(III) without added Fe(II) or synthetic amorphous Fe(III) oxide after extracted by HCl or Tamm's reagent, Fe(II)_a or Fe(III)_a represent the amount of added Fe(II) or synthetic amorphous Fe(III) oxide.

2.5. Analyses

2.5.1. Physicochemical characteristics of MSW samples

The amount of total organic matter of MSW was estimated as loss on ignition (LOI). A sample of approximately 10 g of the material was dried at 105 °C in a ventilated drying box until a constant weight was achieved, the weight noted and then the sample was combusted at 550 °C for 2 h and the LOI was determined. The pH was measured in 1:5 (S/L) suspensions using a pH-meter (DELTA 320). In addition, the BDM was analyzed using the potassium dichromate method [22]. All the analyses were carried out in triplicate to ensure the validity of the results.

2.5.2. Fe(II) and total extractable iron

The Fe(II) extracted by either HCl or Tamm's reagent was quantified photometrically at 510 nm wavelength after chelation with 2 mM *o*-Phenanthroline in 0.7 M sodium acetate buffer, pH 5, in a test volume of 1 ml [27]. The total extractable iron extracted by HCl or Tamm's reagent were determined by flame atomic absorption spectrophotometer AAS (Thermo ice-3500) under optimized measurement conditions. Fe(III) was calculated as the difference between total extractable iron and Fe(II). Standards for the *o*-Phenanthroline assay were made with Fe(NH₄)₂(SO₄)₂·6H₂O. All the analyses were carried out in triplicate to ensure the validity of the results, and all the results of chemical analyses were calculated on a dry-weight basis.

2.5.3. Statistical analysis

In the present study, all the factors selected were examined by orthogonal array L₁₆ (4⁵) and L₉ (3⁴) test design for HCl and Tamm's

reagent extraction. Firstly, according to the analysis of extreme difference (ANEDI), the values of K and R were calculated and presented in Tables 2 and 4. In addition, in order to conclude from the result how much variation each factor causes relative to the total variation observed and whether the difference among the levels of each factor was statistical significant or not, statistical method was also adopted. The extraction results of iron for all sixteen and nine experiments were fed in statistical software SPSS 16.0 for the analysis of variance (ANOVA) for HCl and Tamm's extraction, respectively.

3. Results and discussion

3.1. The physicochemical characteristics of MSW samples

In landfill, MSW were deposited in different layers at different time. Therefore, the basic physicochemical characteristics of MSW in landfill were affected by not only seasons but also the degradation degree of landfill layers. The organic matter content, determined as LOI, pH, moisture content and BDM were presented in Table 5. The six MSW samples, with different deposited ages, represented the stabilization process of landfill. From 0 to 2 years, the landfill experienced two phases, namely the aerobic phase and the anaerobic acid phase. After the burial of refuse, oxygen present in the void spaces of the freshly buried refuse was rapidly consumed in a few days, which was called aerobic phase. As oxygen sources were depleted, the waste became anaerobic, which supported fermentation reactions, and went into the acid phase. During this phase (S_0 to S_2), cellulose and hemicellulose, which comprise 45–60% of the dry weight of MSW and its major biodegradable constituents [3], were decomposed into low molecular dissolved organic matter such as fatty acid. Accordingly, the organic matter content and BDM decreased from $72.40 \pm 1.28\%$ and $54.90 \pm 0.17\%$ to $28.10 \pm 0.93\%$ and $12.40 \pm 0.52\%$, respectively. The moisture content of the refuse also decreased from more than 60% to nearly 40%. After the burial of 2 years, the organic acids that accumulated in the acid phase were converted to methane and carbon dioxide, and experienced the initial and stable methanogenic phase [28]. During the stable phase (S_2 – S_8), the landfill cell turned into a process of slow degradation and the organic matter content and BDM in the refuse decreased slightly from $28.10 \pm 0.93\%$ and $12.40 \pm 0.52\%$ to $15.20 \pm 2.93\%$ and $3.34 \pm 0.74\%$ in 6 years, while the pH and moisture content kept within the range of 7.57 ± 0.04 – 7.72 ± 0.01 and 33.30 ± 2.24 – $42.20 \pm 0.46\%$, respectively. However, as was mentioned above, the rate of refuse decomposition was significantly affected by several factors, such as the initial composition of the refuse, the depth of layers and environmental conditions. Therefore, the time required for different phase would also be varied from one landfill to another. On the other hand, the degradation degree of landfill layers and cells also affected the amount and redox state of iron. The characteristics of sulfides (especially S^{2-}) and humic substances in MSW with different landfill ages varied very much [22,28,29]. As to S^{2-} , we have to bear in mind that it has

a stronger ability to form sulfides with iron than other heavy metals. In addition, ferric iron oxides can be also reduced by hydrogen sulfide, which is a common end product of the anaerobic decomposition of MSW either from sulfur-containing amino acids or by reduction of sulfate [30]. The humic substances can not only affect the mobility but also influence the redox state of iron. Bozkurt et al. [31] demonstrated that humic substances had the strong binding capacity on iron in solid waste. Humic substances are complex organic polymers with redox-active moieties, and thus can chemically reduce ferric iron oxides [32]. Moreover, humic substances (especially humic acid) can act as electron shuttles and affect the microbial iron reduction process [33]. Therefore, the amount and redox state of iron would be varied in different landfill aged MSW.

3.2. Optimized iron extraction techniques

3.2.1. Hydrochloric acid

Although the maximum yield of Fe(II), Fe(III) and total extractable iron were 2530, 3840, and 5950 mg kg^{-1} respectively, the corresponding extraction conditions will not be chosen as the best technique. According to the ANEDI, the values of K and R were calculated by the statistical software and presented Table 2. The factors which influence the yield (mg kg^{-1}) of Fe(II), Fe(III) and total extractable iron were listed in a decreasing order as follows: $B > A > C > E > D$, $B > A > D > E > C$ and $B > A > D > C > E$ in view of the R value. In addition, according to the values of K, we can obtain the maximum yield (mg kg^{-1}) of Fe(II) when the HCl concentrations, liquid-to-solid ratio, extraction time, extraction temperature and shaking speed were $B_4A_4C_4E_3D_3$ (1.00 M, 100, 60 min, 35 °C and 175 rpm). While the maximum yield (mg kg^{-1}) of Fe(III) and total extractable iron can be obtained when the set of $B_4A_3D_4E_1C_4$ (1.00 M, 75, 200 rpm, 35 °C and 60 min) or $B_4A_3D_4C_4E_3$ (1.00 M, 75, 60 min, 200 rpm, and 35 °C) were selected respectively. Although the order of impact factor on the extraction of iron and two better sets were obtained, whether the difference among the levels of each factor was statistical significant or not was still unclear. From the results of ANOVA (Table 6), it was concluded that all the five factors had significant impact on the yield of Fe(III) and total extractable iron, while the yield of Fe(II) was significantly affected by all factors except for shaking speed. According to the analysis by ANEDI and ANOVA, the optimal HCl extraction method was $B_4A_4C_4D_4E_3$ (1.00 M, 100, 60 min, 200 rpm and 35 °C). Through confirmatory test, we got the high yield of Fe(II) and total extractable iron, with a yield of 2750 ± 63 and $6300 \pm 34 \text{ mg kg}^{-1}$, respectively. Although the yield of Fe(III) was $3560 \pm 42 \text{ mg kg}^{-1}$, which still equals to 93% of the maximum yield (3840 mg kg^{-1}), we concluded that the set of $B_4A_4C_4D_4E_3$ was sufficient to extract the iron from MSW and it would be further verified by the fortified recovery followed. The conditions chosen for extracting iron from MSW were in some aspects different from conditions proposed for soils, sediments and activated sludge. In view of higher heterogeneity of MSW samples than soil, sediment and activated sludge and for the sake of getting

Table 5
Basic physicochemical characteristics of MSW samples studied.

Sample	MSW sources	pH	Moisture content ^a (%)	Loss on ignition ^b (%)	Biodegradable matter ^c (%)
S_0	0 year aged refuse	6.17 ± 0.05	52.20 ± 1.16	72.40 ± 1.28	54.90 ± 0.17
$S_{0.5}$	0.5 year aged refuse	8.71 ± 0.05	69.50 ± 0.02	43.60 ± 0.02	23.40 ± 1.14
S_1	1 year aged refuse	7.98 ± 0.03	54.00 ± 0.87	29.80 ± 2.94	10.70 ± 0.20
S_2	2 year aged refuse	7.62 ± 0.04	39.60 ± 1.63	28.10 ± 0.93	12.40 ± 0.52
S_4	4 year aged refuse	7.57 ± 0.04	42.20 ± 0.46	18.00 ± 0.28	9.39 ± 1.16
S_8	8 year aged refuse	7.72 ± 0.01	33.30 ± 2.24	15.20 ± 2.93	3.34 ± 0.74

^a Obtained by drying to constant mass at 105 °C.

^b Obtained by combustion at 550 °C.

^c Obtained by potassium dichromate digestion at room temperature for 12 h.

Table 6
Analysis of variance of the orthogonal experiment for the extraction of iron by HCl.

Design variables	Sum of squares (S_0)			(df) ^a	S^2			F-ratio (F)			P					
	Total				Iron ^b			Fe(II)			Fe(III)			Total		
	Fe(II)	Fe(III)	Total		Fe(II)	Fe(III)	Total	Fe(II)	Fe(III)	Total	Fe(II)	Fe(III)	Total	Fe(II)	Fe(III)	Total
A	2,480,000	5,740,000	15,300,000	3	825,000	1,910,000	5,090,000	77.50	51.50	172.00	0.0001	0.0001	0.0001	0.0001	0.0001	
B	757,000	8,100,000	31,200,000	3	2,520,000	2,700,000	10,400,000	237.00	72.80	353.00	0.0001	0.0001	0.0001	0.0001	0.0001	
C	810,000	1,050,000	3,540,000	3	270,000	350,000	1,180,000	25.40	9.43	40.00	0.0001	0.0001	0.0001	0.0001	0.0001	
D	31,200	5,310,000	4,550,000	3	10,400	1,770,000	1,520,000	0.980	47.70	51.40	0.4160	0.0001	0.0001	0.0001	0.0001	
E	1,180,000	950,000	2,120,000	3	394,000	316,000	708,000	37.10	8.53	24.00	0.0001	0.0003	0.0001	0.0001	0.0001	
Error	341,000	1,190,000	944,000	32	10,600	37,100	29,500									
Total	12,400,000	2,230,000	57,600,000	47	403,000	7,090,000	18,900,000									

^a Refers to the degree of freedom.

^b Refers to Fe(II), Fe(III) and total extractable iron.

representative samples, 1 g instead of 0.1 g sample was chosen. In addition, HCl concentration and liquid-to-solid ratio were higher and extraction time was the same or longer than reported. 0.5 M HCl and 0.1 g sample in 5 ml were commonly chosen by others, and 15 min, 30 min and 1 h was commonly used [11,21,24,25,27,34,35].

3.2.2. Acid ammonium oxalate solution

According to the ANEDI, the values of K and R were calculated by the statistical software and presented in Table 4. The factors influence the yield (mg kg^{-1}) of Fe(II), Fe(III) and total extractable iron were listed in a decreasing order as follows: $C > D > A > E$, $A > E > D > C$ and $A > C > D > E$ in view of the R value. In addition, according to the values of K, we can obtain the maximum yield (mg kg^{-1}) of Fe(II) when the Tamm's reagent liquid-to-solid ratio, extraction time, extraction temperature and shaking speed were $C_3D_3A_3E_1$ (24 h, 175 rpm, 100 and 25°C). While the maximum yield (mg kg^{-1}) of Fe(III) and total extractable iron can be obtained when the set of $A_3E_2D_2C_3$ (100, 30°C , 150 rpm, and 24 h) or $A_3C_3D_2E_2$ (100, 24 h, 150 rpm, and 30°C) were selected respectively. From the results of ANOVA (Table 7), it was concluded that all the four factors had significant impact on the yield of total extractable iron, while the yield of Fe(II) was significantly affected by all factors except for extraction temperature. As to Fe(III), it was only significantly affected by the factor of liquid-to-solid ratio and extraction temperature. Based on the analysis by ANEDI and ANOVA, we got the optimum technique as follows: $A_3C_3D_3E_2$ (100, 24 h, 175 rpm and 30°C). However, the extension of time from 12 to 24 h did not increase the yield of total extractable iron, and the yield of Fe(II) and Fe(III) extracted in 24 h was 4.38% and 0.63% higher than in 12 h respectively. Therefore, an extraction time of 12 h was sufficient for the extraction of Fe(II), Fe(III) and total extractable iron, which was shorter than the condition proposed for soil (4 h) [18]. Through confirmatory test, we got the high yield of Fe(II), Fe(III) and total extractable iron, with a yield of 2480 ± 210 , 3400 ± 395 and $5880 \pm 185 \text{ mg kg}^{-1}$, respectively. Thus, we concluded that the set of $A_3C_2D_3E_2$ was sufficient to extract the iron from MSW and it would be also further verified by the fortified recovery followed.

3.3. Recovery of Fe(II) and Fe(III) in different aged refuse

The recoveries of added Fe(II) or Fe(III) in different deposited aged refuse after extraction with HCl and Tamm's reagent are presented in Table 8. As can be seen from the table, most of the added Fe(II) or Fe(III) can be recovered from different aged refuse after extraction with HCl or Tamm's reagent, demonstrating that both of the extractants were available for iron speciation in MSW. The added Fe(II) in S_1 , S_2 and S_8 were fully recovered with HCl extraction, and full recovery of added Fe(II) was also obtained for S_0 , S_4 and S_8 after extraction with Tamm's reagent. In addition, $95.60 \pm 1.90\%$ and $94.10 \pm 4.20\%$ of the added Fe(II) in S_0 and S_4 were recovered with HCl extraction, while Tamm's reagent extracted $94.00 \pm 5.20\%$ and $93.10 \pm 9.50\%$ of added Fe(II) in S_2 and S_4 . With HCl extraction, the recovery rates of the added Fe(II) in all the MSW samples (except for S_0) were all higher than the recovery rate with Tamm's reagent, which might be attributed to the oxidation of added Fe(II) during Tamm's reagent extraction. Rasmussen and Nielsen [21] observed that oxidation of Fe(II) to Fe(III) took place during the Tamm's reagent extraction, which led to an underestimation of ferrous iron. Although they did not investigate the possible mechanisms for the oxidation, the intrusion of oxygen during extraction might play an important role. Thus, special care must be taken to avoid oxygen contamination during the extraction process.

The analogous results are true for the recovery of Fe(III) after extraction with HCl or Tamm's reagent. With Tamm's reagent, full

Table 7
Analysis of variance of the orthogonal experiment for the extraction of iron by Tamm's reagent.

Design variables	Sum of squares (S_2)		$(df)^a$		S^2		F-ratio (F)		P					
	Fe(II)	Fe(III)	Total	Iron ^b	Iron ^b	Fe(II)	Fe(III)	Total	Fe(II)	Fe(III)	Total			
A	293,000	404,000	6,440,000	2	2	146,000	2,020,000	3,220,000	14.70	80.80	202.00	0.0001	0.0001	0.0001
C	482,000	2130	530,000	2	2	240,000	1060	265,000	24.20	0.04	16.60	0.0001	0.9580	0.0001
D	405,000	129,000	274,000	2	2	202,000	64,700	137,000	20.30	2.60	8.59	0.0001	0.1030	0.0024
E	53,400	48,000	214,000	2	2	26,700	240,000	107,000	2.68	9.60	6.69	0.0960	0.0015	0.0067
Error	180,000	450,000	287,000	18	18	9980	25,000	16,000						
Total	1,410,000	5,100,000	7,750,000	26	26	626,000	2,350,000	3,740,000						

^a Refers to the degree of freedom.

^b Refers to Fe(II), Fe(III) and total extractable iron.

recovery of added Fe(III) were obtained for all the five aged refuse except for S_4 , the recovery of which was also up to $97.80 \pm 6.30\%$. With HCl extraction, the recovery rates of the added Fe(III) in all the aged refuse were slight lower than the recovery rate with Tamm's reagent. The lower recovery rate might be resulted from the reduction of Fe(III) to Fe(II) during extraction in hydrochloric acid. Fe(III), particularly under acid conditions, is susceptible to be reduced by both chemical and biological source, the former of which includes organic compounds (such as formate, citrate, pyruvate and fulvic acid) and inorganic sulfide compounds [17,21,36]. In the anaerobically stored refuse, these types of compounds may be exist, and could be responsible for the reduction of Fe(III) to Fe(II). In our experiment, reduction of Fe(III) did not exert a huge impact on the recovery rate (ca. 3.60–8.10%), thus the reduction of Fe(III) was not a problem during HCl extraction. Above all, the fortified recovery further verified that both of the optimized extraction technique for HCl and Tamm's reagent were suitable for extraction of iron from different deposited aged refuse. Furthermore, microwave power and ultrasound have been proved to be extremely useful auxiliary factor to accelerate sequential extraction procedure [37–39]. Therefore, we consider that these two auxiliary tools might also increase the recover rate of iron extraction processes for HCl and Tamm's reagent extraction, and we'll investigate and present such extraction procedures in our future research in detail.

3.4. Pools of iron in MSW with different deposit ages

With the extractant of HCl, the yield of Fe(II) and total extractable iron increased at an increasing of deposited age and 51.90 ± 0.38 – $83.00 \pm 3.69\%$ was present as Fe(II) (Fig. 1(A)). In the fresh refuse (S_0), the yield of Fe(II) was $2090 \pm 73 \text{ mg kg}^{-1}$ which accounted for 51.90 ± 0.38% of the total extractable iron. However, after 8 years deposit, the yield of Fe(II) and total extractable iron increased up to $9560 \pm 274 \text{ mg kg}^{-1}$ and $13,300 \pm 264 \text{ mg kg}^{-1}$ respectively, and the ratio of Fe(II) to total extractable iron was as high as $71.60 \pm 1.42\%$. In other three aged refuse samples (S_1 to S_4), the elevated amount of Fe(II) was also concomitant with high yield

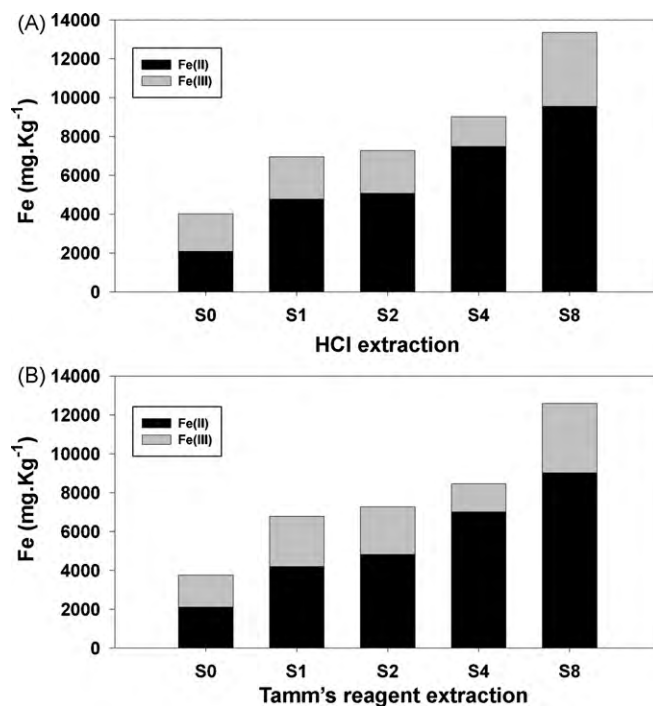


Fig. 1. Pools of iron in MSW with different deposit ages extracted by the extractant of HCl (A) and Tamm's reagent (B).

Table 8

Recoveries of added Fe(II) or Fe(III) in different deposited aged refuse after extraction with HCl and Tamm's reagent.

Sample	Additions (μg)		Recovered (μg)				Recovery rate (%)			
	Fe(II)	Fe(III)	[Fe(II) _y -Fe(II) _n] \pm SD		[Fe(III) _y -Fe(III) _n] \pm SD		Fe(II) \pm SD		Fe(III) \pm SD	
			HCl	Tamm's	HCl	Tamm's	HCl	Tamm's	HCl	Tamm's
S ₀	2240	–	2070 \pm 132	2170 \pm 101	–	–	95.6 \pm 1.9	96.7 \pm 4.5	–	–
	–	2240	–	–	2080 \pm 28	2270 \pm 90	–	–	92.8 \pm 1.2	101.0 \pm 4.0
S ₁	5600	–	5640 \pm 265	5290 \pm 86	–	–	101.0 \pm 4.7	94.5 \pm 1.5	–	–
	–	2240	–	–	2160 \pm 53	2320 \pm 194	–	–	96.4 \pm 2.4	104.0 \pm 3.5
S ₂	5600	–	5440 \pm 351	5260 \pm 292	–	–	97.2 \pm 9.6	94.0 \pm 5.2	–	–
	–	2240	–	–	2060 \pm 52	2190 \pm 142	–	–	91.9 \pm 2.3	97.8 \pm 6.3
S ₄	8400	–	7910 \pm 351	7820 \pm 800	–	–	94.1 \pm 4.2	93.1 \pm 9.5	–	–
	–	2240	–	–	2130 \pm 85	2220 \pm 36	–	–	95.3 \pm 3.8	99.5 \pm 1.6
S ₈	11,200	–	11,600 \pm 944	11,300 \pm 219	–	–	103.0 \pm 8.4	101.0 \pm 2.0	–	–
	–	5590	–	–	540 \pm 198	5780 \pm 465	–	–	93.7 \pm 3.5	103.0 \pm 8.3

of total extractable iron, and the ratio of Fe(II) to total extractable iron varied from $68.70 \pm 1.31\%$ to $83.00 \pm 3.69\%$. As was speculated above, extractable of iron varied very much in MSW with different deposit ages. After 8 years deposit, the amount of total extractable iron was more than three times as high as that in the fresh refuse. This discrepancy may be ascribed to the difference of the degradation degree of MSW with different deposit ages. Investigations have shown that less than 0.02% of heavy metals received at landfills are leached from the landfill after 30 years [3,40]. Most of the heavy metals dissolved by leachate were immobilized again by adsorption and precipitation during the leaching in MSW landfill. As was mentioned above, the parameters (e.g., CEC and humus) relevant with heavy metals retention varied in MSW with different landfill ages. Especially when the landfill reaches the humic phase, a minor fraction of the original organic matter remains and it is in the form of humic substances, which have a strong binding capacity on heavy metals [29]. Therefore, iron contents in MSW were increased relatively other than decreased. That is to say, the rate of iron released by leachate from MSW landfill was much lower than the rate of organic matter degradation and then its level (percentage) was increased relatively in spite of minor portion of iron leaching from the landfill.

With the extractant of Tamm's reagent, the yield of Fe(II) and total extractable iron also increased at an increasing of deposited age (Fig. 1(B)). In the fresh refuse (S₀), no significant difference of Fe(II) was found extracted with both HCl and Tamm's reagent, while with Tamm's reagent the yield of total extractable iron was $3780 \pm 94 \text{ mg kg}^{-1}$, which is lower than the amount of total extractable iron yielded by HCl. In the other four aged refuse samples (S₁ to S₈), the yield of total extractable iron extracted by Tamm's reagent were slightly lower than those by HCl and no statistically significant difference was found for the yield total extractable iron extracted by the two extractants. However, with the extractant of Tamm's reagent, the yields of Fe(II) were lower than those extracted by HCl and the differences only varied from 263 mg to 577 mg for 1 kg samples. As was mentioned above, the waste loading rate is about 1940–4000 t day⁻¹, thus the yield of Fe(II) extracted by Tamm's reagent will be up to 510 kg to 2310 kg, less than those extracted by HCl for 1 day's loading capacity. Therefore, we can conclude that minor difference between two extraction procedures will exert a huge impact on the research of iron cycling with carbon, sulfur and nitrogen cycles from the angle of the landfill. In addition, even though the anaerobic techniques that are required in the Tamm's reagent extraction are not difficult or expensive and are routinely used in a large number of microbiological laboratories, it is laborious and much more time-consuming than HCl extraction, thus the latter one might be a good choice for the evaluation of the redox state of iron in the MSW.

4. Conclusions

Excavation of MSW and optimization of extraction procedure by orthogonal test design supplied a promising tool for speciation extractable iron into ferric and ferrous, and investigating the role of iron played on the migration and transformation of inorganic and organic pollutants in the MSW. The conditions chosen for extracting iron from MSW with both the extractant of HCl and Tamm's reagent were in some aspects different from conditions proposed for soils, sediments and activated sludge. The concentration of HCl and liquid-to-solid ratio was a little higher, while the extraction time with HCl and Tamm's reagent were the same or longer than reported. Specified with two extractants under optimized conditions showed that extractable iron increased at an increasing of deposited age and 60–83% was present as Fe(II) in the aged MSW.

Although the reduction and oxidation of iron which leads to an overestimation or underestimation of the Fe(II) pool in the MSW probably took place during extraction in HCl or Tamm's reagent, both methods described here may improve understanding of the pool of iron in the different deposited MSW. However, even though the anaerobic techniques that are required in the Tamm's reagent extraction are not difficult or expensive and are routinely used in a large number of microbiological laboratories, it is laborious and much more time-consuming than HCl extraction, thus the latter one might be a good choice for the evaluation of the redox state of iron in the MSW.

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